UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER OF PATENTS AND TRADEMARKS
Washington, D.C. 20231
www.uspto.gov

APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
09/463,494	07/25/2000	MANFRED T. REETZ	STUDIEN-268-	6396

7590 10/02/2002

NORRIS McLAUGHLIN & MARCUS, P.A.
220 EAST 42nd STREET
30th FLOOR
NEW YORK, NY 10017

EXAMINER
PATTERSON, CHARLES L JR

ART UNIT	PAPER NUMBER
1652	

DATE MAILED: 10/02/2002

Please find below and/or attached an Office communication concerning this application or proceeding.

Office Action Summary	Application No.	Applicant(s)	
	09/463,494	REETZ ET AL.	
	Examiner	Art Unit	
	Charles L. Patterson, Jr.	1652	

-- Th MAILING DATE of this communication app ars on the cov r she t with the correspond nce address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If the period for reply specified above is less than thirty (30) days, a reply within the statutory minimum of thirty (30) days will be considered timely.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133).
- Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *7/25/00, 2/25/02 & 8/19/02* .

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *36-38* is/are pending in the application.

4a) Of the above claim(s) _____ is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *36-38* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☒ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☒ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

11)☐ The proposed drawing correction filed on _____ is: a)☐ approved b)☐ disapproved by the Examiner.

If approved, corrected drawings are required in reply to this Office action.

12)☐ The oath or declaration is objected to by the Examiner.

Priority under 35 U.S.C. §§ 119 and 120

13)☒ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☒ All b)☐ Some * c)☐ None of:

1.☒ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____ .

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

14)☐ Acknowledgment is made of a claim for domestic priority under 35 U.S.C. § 119(e) (to a provisional application).

a) ☐ The translation of the foreign language provisional application has been received.

15)☐ Acknowledgment is made of a claim for domestic priority under 35 U.S.C. §§ 120 and/or 121.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☒ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☐ Information Disclosure Statement(s) (PTO-1449) Paper No(s) _____ .
4) ☐ Interview Summary (PTO-413) Paper No(s). _____ .
5) ☐ Notice of Informal Patent Application (PTO-152)
6) ☐ Other: .

The preliminary amendment filed 7/25/00 directs the cancellation of the original claims and the substitution of other claims, 1-3. There are 35 original claims and therefore the new claims have been re-numbered as 36-38 is accord with 37 CFR 1/126. These new numbers are used henceforth.

Claim 36 is rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

Claim 36 is indefinite in the recitation of "modified" on line 3. How is the PCR to be modified? The claim is also indefinite in the recitation of "the amplified DNA" in line 4. There is no antecedent basis previously in the claim for this term.

Finally, claim 36 is indefinite in the recitation of "optionally" on line 7. It is unclear whether the limitation is meant to be limiting or simply illustrative.

The following is a quotation of the first paragraph of 35 U.S.C. 112:

> The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

Claims 36-38 are rejected under 35 U.S.C. 112, first paragraph, as containing subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventor(s), at the time the application was filed, had possession of the claimed invention and in such a way as to enable one skilled in the art to which it pertains, or with which it is most nearly connected, to make and/or

use the invention. This is a combination written description and enablement rejection.

Claim 36 is drawn to preparing hydrolase mutants having "improved properties with respect to stereo- or regioselectivity" by using a modified PCR reaction where the mutation rate and number of mutations in "the amplified DNA" [see 35 USC § 112 second paragraph rejection *supra*] is adjusted by adjusting the concentrations of Mg^{2+}, Mn^{2+}, or the deoxynucleotides and by adjusting the number of cycles. The claim also contains the optional step of fragmenting the genes and reassembly of the fragments. To start with, the process of the instant specification does not preferentially produce "hydrolase mutants having improved properties with respect to stereo- or regioselectivity", but rather causes random mutagenesis that is then assayed to see whether mutants have been produced having improved enantioselectivity. Furthermore, the concentrations of Mg^{2+}, Mn^{2+}, or the deoxynucleotides do not appear to have been varied.

In the paragraph spanning pages 8-9 it is taught that the method of the instant invention first uses DnaseI to cleave the gene into fragments and then the fragment are mutagenized by using the conditions of conventional PCR *in vitro* without adding any PCR primers. None of this is in the instant claims. Also, this does not appear to be the case here even though the specification on page 8-9 states that it is because in the last paragraph of page 21 it is stated that "10 pmol each of the primers" is used in the "mutagenic PCR" procedure.

Furthermore, it is not understood how the fragments are recombined (annealed). On page 9 it is stated that "hybridization occurs of sequence-homologous fragments which may be derived from different mutated lipase or esterase genes" but exactly how this is done is not stated in the specifica-

tion. In describing example 1 on page 21, last paragraph it is stated that "[a]fter purification of the PCR product using a Qiagen Qiaquick Column®, it serves as a template in a mutagenic PCR". It is not clear from this recitation exactly what serves as a template for the mutagenic PCR. Is the sentence meant to say that the column is a template?

Claim 36(b) states that the mutated genes of step (a) or mixtures of unmutated genes and mutated genes are mutagenized by "enzymatically fragmenting said genes, followed by enzymatic reassembly of the fragments produced to give complete recombinant hydroxylase genes". It is not seen how this step will mutate the genes.

Figure 1 is stated on page 18 to show "the experimentally obtained measured curves for the determination of the apparent enantioselectivity (E_{app}) in the hydrolysis of (R)- and (S)-2-methyldecanoic acid p-nitrophenyl ester with" certain lipase mutants. Nothing else is apparently stated about this figure in the specification. It is presumed that the increase in OD shown in the figure is indicative of the increase of the p-nitrophenyl radical due to hydrolysis with time, but this is not stated. There are two curves shown in each section of Fig. 1, but there is no apparent statement in the specification as to what these two curves represent.

It is maintained that one of ordinary skill in the art would not be taught how to make "hydrolase mutants having improved properties with respect to stereo- or regioselectivity" as claimed. Furthermore, it would not appear to one of ordinary skill that applicant was in possession of the claimed method when the application was filed.

The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

Claims 36-38 are rejected under 35 U.S.C. 103(a) as being unpatentable over either Williams, et al. (A), Zhou, et al. (U), Leung, et al. (V), Cadwell, et al. (W) or Shinkai, et al. (X).

Williams, et al. teach in column 1, line 36-52 that random mutations can be generated using PCR and that in an improved version of this method the fidelity of the DNA synthesis by Taq polymerase can be further reduced by adding Mn, Mg and the relevant dNTPs.

Zhou, et al (U) teach that Taq polymerase lacks a 3′→ 5′ editing activity and that random mutagenesis will occur with short DNA molecules using PCR with Mn. They use 4 steps: PCR, digestion with restriction enzymes at each end, ligation into a restriction enzyme digested vector and introduction of the resulting recombinant DNA into cells.

Leung, et al. (V) teach that random point mutations can be made to DNA by using PCR conditions that reduce the fidelity of DNA synthesis by Taq polymerase and inserting the mutated DNA fragments into cloning vectors to generate random mutants.

Cadwell, et al. (W) teach that mutations can be made in DNA by using PCR with Taq polymerase. They state in the paragraph spanning column 2 and 3 on page 29 that increased Taq polymerase concentration, increased extension time, increased concentration of $MgCl_2$, addition of $MnCl_2$ and increased concentration of dNTPs will increase mutagenicity.

Shinkai, et al. teach the production of lipase gene mutants by using "error-prone PCR" carried out according to the method taught by Cadwell, et al. (W).

It would have been obvious to one of ordinary skill in the art to produce mutants using PCR as taught in the instant references. The rate of mutation could have been varied by varying the concentrations of Mn, Mg and dNTPs, as taught in the instant references. The type of mutation produced could have been evaluated by assaying different substrates under different conditions. It would have been further obvious that if more mutation was desired the product of the first mutagenic PCR could be used as the starting material for another round of PCR.

Copies of Leung, et al., Cadwell, et al. and Shinkai, et al. are not being sent because they were cited in the PCR search report.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Charles L. Patterson, Jr., PhD, whose telephone number is 703-308-1834. The examiner can normally be reached on Monday - Friday, 7:30-4:00.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Ponnathapura Achutamurthy can be reached on 703-308-3804. The fax phone numbers for the organization where this application or proceeding is assigned are 703-308-4242 for regular communications and 703-308-0294 for After Final communications.

Any inquiry of a general nature or relating to the status of this application or proceeding should be directed to the receptionist whose telephone number is 703-308-0196.

Charles L. Patterson, Jr.
Primary Examiner
Art Unit 1652

Patterson
September 30, 2002